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Sources and Movement of *Salmonella* through Integrated Poultry Operations: A Multistate Epidemiological Investigation

J. S. BAILEY,* N. J. STERN, P. FEDORKA-CRAY, S. E. CRAVEN, N. A. COX, D. E. COSBY, S. LADELY, AND M. T. MUSGROVE

Richard B. Russell Agricultural Research Center, Poultry Microbiological Safety Research Unit, Agricultural Research Service, U.S. Department of Agriculture, P.O. Box 5677, Athens, Georgia 30604-5677, USA

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ABSTRACT

The prevalence of *Salmonella* from numerous sources in 32 integrated broiler operations of high- and low-performing broiler houses was characterized from four states across four seasons. Previous studies of *Salmonella* in broilers have been limited in scope, offering only a snapshot of pathogen prevalence as seen on a small number of individual farms. Twenty-six different sample types were collected from the hatchery to the end of processing, and *Salmonella* was found in all sample types. A total of 10,740 samples were analyzed for *Salmonella,* and 973 (9.1%) of these samples, including 49 of 798 (6.1%) carcass rinse samples, were *Salmonella* positive. Hatchery transport pads (389 of 765, 50.8%), flies (28 of 150, 18.7%), drag swabs (57 of 402, 14.2%), and boot swabs (20 of 167, 12%) were samples from which *Salmonella* was most frequently isolated. Thirty-six different serotypes were identified, and the most frequently encountered serotypes were *Salmonella* Senftenberg, *Salmonella* Thompson, and *Salmonella* Montevideo. Determining critical contaminating sources and following the movement of *Salmonella* through integrated poultry operations will help researchers and the industry develop practical intervention strategies.

Control of *Salmonella* is complicated, because there are numerous potential sources of *Salmonella* contamination in an integrated poultry operation, including chicks, feed, rodents, wild birds, insects, transportation, farm environment, and processing plant environment. All sources of *Salmonella* are potentially important, but it is important to be able to characterize the relative importance of the different sources under specific management and environmental conditions. There are many factors that can influence the relative importance of various sources of *Salmonella;* these include (i) age of the chicken; (ii) survival through the gastric barrier; (iii) competing bacteria in the intestinal tract; (iv) availability of a hospitable colonization site; (v) nature of diet; (vi) physiological status of the chicken; (vii) health and disease status of the chicken; and (viii) medication effects, which will influence the potential colonization of chickens with *Salmonella.*

Previous studies *(1, 7, 13)* have concluded that the hatchery may be the most important source of contamination, regardless of grow-out conditions, for two reasons: (i) the newly hatched chick is more susceptible to colonization than older birds, and (ii) chicks are often exposed to *Salmonella* in the hatchery. Milner and Shaffer *(17)* first observed that colonization of chicks was dose-dependent and varied with day of challenge. They found that 1-day-old chicks could be colonized with less than five cells of *Salmonella* and that later colonization was irregular and required higher doses of *Salmonella.* Cox et al. *(9)* found that after cloacal challenge, chicks were colonized with only two cells of *Salmonella.* Two-week-old chicks have a mature gut microflora *(6)* and are thus more resistant to intestinal colonization by salmonellae. Bailey et al. *(2, 4)* demonstrated that a single *Salmonella*-contaminated egg could substantially contaminate other eggs and chicks in a hatching cabinet. Cox et al. *(8, 10)* showed substantial *Salmonella* contamination of egg fragments, belting material, and paper pads in commercial hatcheries, indicating many opportunities for contamination of newly hatched chicks. Two studies looked at commercial hatcheries and found 5 to 9% of 1-day-old chicks to be salmonellae positive *(15, 16).*

Other researchers have concluded that the grow-out environment may be the most important source of the *Salmonella* serotypes found at slaughter. Lahellec and Colin *(16)* observed that *Salmonella* serotypes originating in the hatchery were less important in the final product than those present in the house or those introduced into the house by vectors during rearing. Bailey et al. *(3)* and Blankenship et al. *(7)* found that the environment was the primary source of contaminating *Salmonella* in chicken houses not treated with competitive exclusion microflora.

Other sources have been implicated in the proliferation and spread of *Salmonella* in the chicken industry. Erwin *(12)* first recovered viable *Salmonella* from commercial poultry feed, and since that time, the role of feed and feed ingredients in the spread of *Salmonella* through the poultry industry has received a great deal of attention. Less than one *Salmonella* per g of feed has been shown to establish colonization in 1- to 7-day-old chicks *(19).* Therefore, under some conditions, feed can be an important source of *Salmonella.* However, others, including Goren et al. *(13)* in

* Author for correspondence. Tel: 706-546-3356; Fax: 706-546-3771; E-mail: jsbailey@saa.ars.usda.gov.

TABLE 1. Salmonella *from all sample types and times for four integrators across all seasons and high- and low-production houses (a total of 32 houses and 8,739 samples)*

Sample	Total	Integrator A	Integrator B	Integrator C	Integrator D
Paper pads	389/765 (50.8%)	65/200 (32.5%)	83/175 (47.4%)	51/192 (26.6%)	190/198 (96.0%)
Feces	168/2,546 (6.6%)	5/598 (0.8%)	77/749 (10.3%)	55/600 (9.2%)	31/599 (5.2%)
Water line	10/731 (1.4%)	0/178 (0.0%)	3/224 (1.3%)	0/140 (0.0%)	7/189 (3.7%)
Water cup	15/797 (1.9%)	1/191 (0.5%)	3/228 (1.3%)	5/186 (2.7%)	6/192 (3.1%)
Litter	84/799 (10.5%)	3/192 (1.6%)	35/228 (15.4%)	28/187 (15.0%)	18/192 (9.4%)
Feed hopper	6/258 (2.3%)	1/62 (1.6%)	3/76 (3.9%)	0/57 (0.0%)	2/63 (3.2%)
Feeder	6/263 (2.3%)	0/64 (0.0%)	3/76 (3.9%)	0/61 (0.0%)	3/62 (4.8%)
Drag swab	57/402 (14.2%)	2/96 (2.1%)	24/114 (21.1%)	16/96 (16.7%)	15/96 (15.6%)
Wall swab	9/268 (3.4%)	2/64 (3.1%)	2/76 (2.6%)	5/64 (7.8%)	0/64 (0.0%)
Fan swab	9/268 (3.4%)	1/64 (1.6%)	1/76 (1.3%)	5/64 (7.8%)	2/64 (3.1%)
Mouse samples	3/49 (6.1%)	2/16 (12.5%)	0/2 (0.0%)	1/27 (3.7%)	0/4 (0.0%)
Wild-bird feces	14/213 (6.6%)	3/49 (6.1%)	5/35 (14.3%)	2/46 (4.3%)	4/83 (4.8%)
Animal feces	3/100 (3.0%)	2/52 (3.8%)	0/6 (0.0%)	0/4 (0.0%)	1/38 (2.6%)
Insects	11/386 (2.8%)	6/98 (6.1%)	1/133 (0.8%)	2/48 (4.2%)	2/107 (1.9%)
Dirt, near entrance	8/131 (6.1%)	2/32 (6.3%)	5/37 (13.5%)	1/30 (3.3%)	0/32 (0.0%)
Standing water	4/79 (5.1%)	1/21 (4.8%)	1/24 (4.2%)	1/12 (8.3%)	1/22 (4.5%)
Boot swab	20/167 (12.0%)	4/28 (14.3%)	7/42 (16.7%)	6/51 (11.8%)	3/46 (6.5%)
Fly strip	28/150 (18.7%)	11/44 (25.0%)	2/38 (5.3%)	8/27 (29.6%)	7/41 (17.1%)
Cecal droppings	16/367 (4.4%)	1/100 (1.0%)	11/120 (9.2%)	3/60 (5.0%)	1/87 (1.1%)
Total	860/8,739 (9.8%)	112/2,149 (5.2%)	266/2,459 (10.8%)	189/1,952 (9.7%)	293/2,179 (13.4%)

a study involving over 8 million broilers, have shown that serotypes of *Salmonella* found on final processed carcasses were found from hatchery samples but not from feed samples. Rodents, particularly mice, have been implicated in the spread of *Salmonella* Enteritidis, particularly in the layer industry *(14)*.

Previous studies of *Salmonella* in poultry have been limited in scope, offering only a snapshot of pathogen prevalence as seen on a small number of individual farms. The objective of this study was to determine the relative importance of all known sources of *Salmonella* from the hatchery through growout and processing in high- and low-production flocks from four integrated operations located in four states across four seasons.

MATERIALS AND METHODS

Flock location and husbandry conditions. The participating companies were located in Alabama, Arkansas, California, and Georgia, respectively, and were randomly coded A through D. Each company provided a rearing house on two different farms—one with a history of high broiler-growth performance, the other associated with low performance. At each of the sites, separate broiler flocks were studied during the spring, summer, fall, and winter of 1998—a total of eight flocks per company. To offset seasonal variability from one part of the country to another, seasons for all locations were defined as "spring": March, April, and May; "summer": June, July, and August; "fall": September, October, and November; and "winter": December, January, and February. Because some of the trials crossed from one season to another, the season was listed as the one in which the majority of the trial took place.

Each broiler flock was comprised of approximately 20,000 birds, involving an "all-in, all-out" stocking policy with a rearing period of 6 weeks for companies A, C, and D and 7 to 8 weeks for company B. Drinking water for the birds was obtained from a chlorinated main supply, a nonchlorinated well, or both. In most cases, the litter on which the birds were kept was fully removed annually, when cleaning and disinfection of the houses were carried out. At other times, the top layer of used litter was removed ("decaking") and replaced with a layer of fresh litter ("top dressing"). By contrast, the houses of producer in state C were emptied and fully cleaned and disinfected between flocks or for every alternate flock. To increase ventilation during hot weather, the houses were partially opened to the outside, and, where required, water sprays were used to provide evaporative cooling.

All producers operated a rodent control program; however, little use was made of disinfectant foot baths, apart from the producer in state C, who also made available clean, dedicated footwear. Most of the farms, except for that of the producer in state C, were close to other livestock, which sometimes had access to the site, while dogs, cats, or both were present on all farms, except for the producer in state C.

Conditions of processing. All the processing plants were modern, highly mechanized operations that processed up to 7,000 carcasses/h. After scalding, plucking, and evisceration, the carcasses were spray washed and chilled in counterflow, water-immersion chillers. The input water to the chiller contained up to 50 mg/liter of free available chlorine.

Sample handling. All samples from both farms and processing plants were collected on each occasion within 1 to 2 h and transferred to insulated boxes containing ice packs for overnight transport to the laboratory. Whenever samples were collected and delivered on the same day, the samples were held under refrigeration overnight to ensure uniformity of sampling among farms. All samples were processed within 36 h of collection.

Sampling at placement. Samples taken at day of placement were paper pads (25), water line swabs (6), water cup/nipple swabs (6), litter (6), feeders (2), feed hoppers (2), wall swabs (2), fan swabs (2), drag swabs (3), insects (2 inside and 2 outside if available), boot swabs (3 if available from farm workers or hatchery personnel), dirt sample (1), standing water (1 if available),

TABLE 2. Salmonella *from all sample types by week of growout from four integrators across all seasons and high- and low-production houses (a total of 32 houses and 8,739 samples)*

Sample	Total		Week 0		Week 2		Week 4		Week 6		Week 8	
Paper pads	389/765	(50.8%)	0/0	(0%)	0/0	(0%)	0/0	(0%)	0/0	(0%)	0/0	(0%)
Feces	168/2,546	(6.6%)	0/0	(0%)	57/798	(7.1%)	20/800	(2.5%)	50/798	(6.3%)	41/150	(27.3%)
Water line	10/731	(1.4%)	7/156	(4.5%)	0/182	(0%)	0/180	(0%)	1/177	(0.6%)	2/36	(5.6%)
Water cup	15/797	(1.9%)	7/191	(3.7%)	4/190	(2.1%)	2/191	(1.0%)	0/189	(0.0%)	2/36	(5.6%)
Litter	84/799	(10.5%)	3/192	(1.6%)	30/191	(15.7%)	13/191	(6.8%)	23/189	(12.2%)	15/36	(41.7%)
Feed hopper	6/258	(2.3%)	1/63	(1.6%)	2/63	(3.2%)	1/63	(1.6%)	0/57	(0.0%)	2/12	(16.7%)
Feeder	6/263	(2.3%)	0/64	(0.0%)	4/64	(6.3%)	0/64	(0.0%)	1/59	(1.7%)	1/12	(8.3%)
Drag swab	57/402	(14.2%)	1/96	(1.0%)	17/96	(17.7%)	13/96	(13.5%)	16/96	(16.7%)	10/18	(55.6%)
Wall swab	9/268	(3.4%)	2/64	(3.1%)	4/64	(6.3%)	3/64	(4.7%)	0/64	(0.0%)	0/12	(0.0%)
Fan swab	9/268	(3.4%)	0/64	(0.0%)	7/64	(10.9%)	2/64	(3.1%)	0/64	(0.0%)	0/12	(0.0%)
Mouse rinse	2/24	(8.3%)	1/5	(20%)	1/8	(12.5%)	0/7	(0.0%)	0/4	(0.0%)	0/0	(ND)
Mouse entrails	1/25	(4.0%)	0/6	(0.0%)	0/8	(0.0%)	1/7	(14.3%)	0/4	(0.0%)	0/0	(ND)
Wild-bird feces	14/213	(6.6%)	5/56	(8.9%)	3/58	(5.2%)	4/47	(8.5%)	0/41	(0.0%)	2/11	(18.2%)
Animal feces	3/100	(3%)	2/29	(6.9%)	0/26	(0.0%)	0/24	(0.0%)	1/21	(4.8%)	0/0	(ND)
Insects	11/386	(2.8%)	4/110	(3.6%)	3/77	(3.9%)	2/89	(2.2%)	1/87	(1.1%)	1/23	(4.3%)
Dirt, near entrance	8/131	(6.1%)	1/32	(3.1%)	3/31	(9.7%)	1/31	(3.2%)	2/32	(6.3%)	1/5	(20.0%)
Standing water	4/79	(5.1%)	2/22	(9.1%)	2/18	(11.1%)	0/19	(0.0%)	0/17	(0.0%)	0/3	(0.0%)
Boot swab	20/167	(12.0%)	6/61	(9.8%)	5/36	(13.9%)	3/31	(9.7%)	4/33	(12.1%)	2/6	(33.3%)
Fly strip	28/150	(18.7%)	0/0	(ND)	13/52	(25.0%)	5/47	(10.6%)	10/46	(21.7%)	0/5	(0.0%)
Cecal droppings	16/367	(4.4%)	0/0	(ND)	7/79	(8.9%)	4/118	(3.4%)	3/140	(2.1%)	2/30	(6.7%)
Total	890/8,739	(9.8%)	431/1,976	(21.8%)	162/2,105	(7.7%)	74/2,133	(3.5%)	112/2,118	(5.3%)	81/407	(19.9%)

TABLE 3. *Transport and in-plant* Salmonella *from four integrators across four seasons*

Season	Sample type	Integrator A	Integrator B	Integrator C	Integrator D
Spring	Carcass rinse	0/50 (0.0%)	6/50 (12.0%)	4/50 (8.0%)	2/50 (4.0%)
	Postchill H_2O	0/10 (0.0%)	2/10 (20.0%)	0/10 (0.0%)	1/10 (10.0%)
	Prescald H_2O	0/0 (ND)	0/10 (0.0%)	0/10 (0.0%)	0/10 (0.0%)
	Postscald H_2O	4/10 (40.0%)	0/10 (0.0%)	0/10 (0.0%)	0/10 (0.0%)
	Pretransport coop	0/0 (ND)	0/20 (0.0%)	0/20 (0.0%)	4/20 (20.0%)
	Posttransport coop	1/20 (5.0%)	6/20 (30.0%)	2/20 (10.0%)	1/20 (5.0%)
Totals[a]		5/100 (5.0%)	14/130 (10.8%)	6/130 (4.6%)	8/130 (6.2%)
Summer	Carcass rinse	0/50 (0.0%)	1/50 (2.0%)	1/50 (2.0%)	1/49 (2.0%)
	Postchill H_2O	0/10 (0.0%)	1/10 (10.0%)	0/10 (0.0%)	0/10 (0.0%)
	Prescald H_2O	0/10 (0.0%)	0/10 (0.0%)	0/5 (0.0%)	0/10 (0.0%)
	Postscald H_2O	5/10 (50.0%)	0/10 (0.0%)	0/10 (0.0%)	0/10 (0.0%)
	Pretransport coop	0/20 (0.0%)	4/20 (20.0%)	0/20 (0.0%)	0/0 (ND)
	Posttransport coop	0/19 (0.0%)	10/20 (50.0%)	0/20 (0.0%)	0/20 (0.0%)
Totals[a]		5/129 (3.9%)	16/130 (12.3%)	1/125 (0.8%)	1/109 (0.9%)
Fall	Carcass rinse	1/50 (2.0%)	18/50 (36.0%)	0/49 (0.0%)	0/50 (0.0%)
	Postchill H_2O	0/10 (0.0%)	0/10 (0.0%)	0/10 (0.0%)	1/10 (10.0%)
	Prescald H_2O	5/10 (50.0%)	0/10 (0.0%)	0/10 (0.0%)	1/10 (10.0%)
	Postscald H_2O	6/10 (60.0%)	0/10 (0.0%)	0/5 (0.0%)	0/10 (0.0%)
	Pretransport coop	0/20 (0.0%)	1/20 (5.0%)	0/10 (0.0%)	3/20 (15.0%)
	Posttransport coop	2/20 (10.0%)	1/20 (5.0%)	0/20 (0.0%)	3/20 (15.0%)
Totals[a]		14/130 (10.8%)	20/130 (15.4%)	0/114 (0.0%)	8/130 (6.2%)
Winter	Carcass rinse	0/50 (0.0%)	0/50 (0.0%)	3/50 (6.0%)	12/50 (24.0%)
	Postchill H_2O	0/10 (0.0%)	0/10 (0.0%)	0/10 (0.0%)	0/10 (0.0%)
	Prescald H_2O	0/10 (0.0%)	0/10 (0.0%)	0/6 (0.0%)	0/10 (0.0%)
	Postscald H_2O	0/10 (0.0%)	0/10 (0.0%)	0/9 (0.0%)	0/10 (0.0%)
	Pretransport coop	0/20 (0.0%)	0/20 (0.0%)	0/20 (0.0%)	0/20 (0.0%)
	Posttransport coop	0/20 (0.0%)	0/20 (0.0%)	0/20 (0.0%)	0/20 (0.0%)
Totals[a]		0/130 (0.0%)	0/130 (0.0%)	3/124 (2.4%)	12/130 (9.2%)
Grand totals[a]		24/489 (4.9%)	50/520 (9.6%)	10/493 (2.0%)	29/499 (5.8%)

[a] Totals include 10 prechill H_2O samples per integrator per season (grand totals by 40 per integrator). All samples were negative for *Salmonella*; data not presented, but numbers are included in the totals and grand totals for statistical purposes.

animal feces (2 if available), and wild-bird feces (4 if available). The paper pads were removed from the transport trays after the birds were put in the house and placed into ziplock bags. The water line and water cup swabs were collected by using four or five sterile cotton-tipped applicators and inserting the applicators into the water line at the ends of the line or by swabbing the drinking nipples or cups, making sure that the swabs were moistened by the water. The applicators were then placed in sterile whirl-pac bags. Litter, feed, feed hoppers, and dirt samples consisted of aseptically collected 50- to 100-g amounts of the dry material placed into a sterile whirl-pac bag. Wall swabs, fan swabs, and boot swabs were collected by premoistening 4 by 4-in. gauze pads with sterile buffered peptone water (BPW; Difco, Becton Dickinson, Sparks, Md.) and then swabbing approximately 100 cm^2 for the wall and fan swabs and both boots of the farm and hatchery personnel. The drag swabs were from a commercial source (Bio-spo, Solar Biologicals, Ogdensburg, N.Y.) and were collected by dragging straight down one side of the house, zigzagging up the middle of the house, and straight down the other side of the house. The drag swabs were then placed into sterile whirl-pac bags. As many insects as possible were collected inside and outside, sorted by the types and origins, and placed into sterile whirl-pac bags. Standing water was collected into sterile 50-ml disposable centrifuge tubes. Animal feces, where available, were collected by placing approximate 50 to 100 g into sterile whirl-pac bags. Wild-bird feces, where available, were collected by placing droppings into sterile whirl-pac bags.

Sampling at weeks 2, 4, 6, and 8. Samples taken at weeks 2, 4, 6, and 8 were fecal droppings (25), cecal droppings (5 if available), rodent samples (2 to 4 if available), water line swabs (6), water cup and nipple swabs (6), litter (6), feeders (2), feed hoppers (2), wall swabs (2), fan swabs (2), drag swabs (3), insects (2 inside and 2 outside if available), fly strips (2) (Quick Strike, Sandos Agro, Inc., Des Plaines, Ill.), boot swabs (3 if available from farm workers), dirt sample (1), standing water (1 if available), animal feces (2 if available), and wild-bird feces (4 if available). Fresh fecal droppings were aseptically collected into 50-ml disposable centrifuge tubes. The cecal droppings were sampled by dipping two or three sterile cotton-tipped applicators into the cecal dropping. The applicators were then placed into sterile whirl-pac bags. The mice were collected using Victor Tin Cat repeating mouse traps (Woodstream Corp., Lititz, Pa.) baited with apple halves and peanut butter. The traps were set no more than 72 h before sample collection. The mice were killed by cervical dislocation and placed into sterile whirl-pac bags before transporting to the laboratory. The fly strips were placed at the time of bird placement by hanging from the ceiling joists with a ziplock bag stapled to the strip for use as a collection bag. The flies were removed from the bags on the appropriate sample day and placed

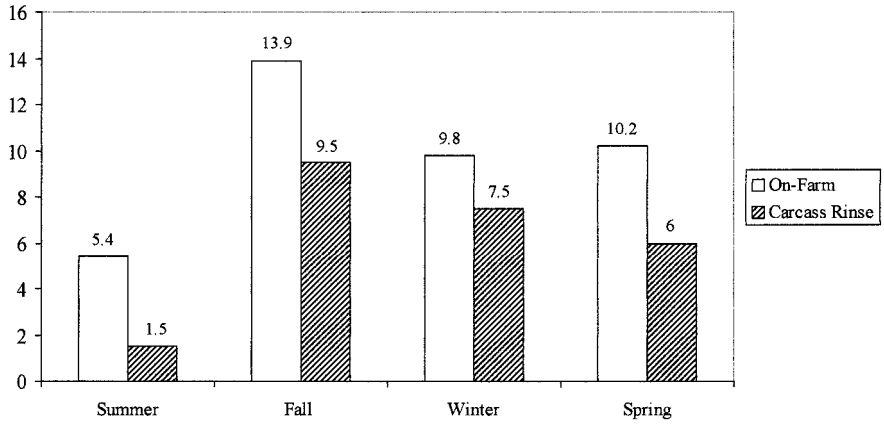

FIGURE 1. *Prevalence of* Salmonella *by season for all sample types and locations. Summer = June, July, and August; fall = September, October, and November; winter = December, January, and February; and spring = March, April, and May.*

into sterile whirl-pac bags. The remaining samples were collected according to the protocol used at placement.

Sampling at processing plants. Samples taken at time of processing were pretransport coop swabs (10), posttransport coop swabs (10), prescald water (5), postscald water (5), prechill water (5), postchill water (5), and postchill carcass rinses (25). The pretransport coop swabs were collected on the farm before loading the coops by swabbing approximately 300 cm^2 of the bottom surface of the transport coops with 4 by 4-in. gauze swabs premoistened with BPW. The swabs were then placed into sterile whirl-pac bags. The posttransport coop swabs were collected at the plant after the birds were removed from the coops using the same protocol as for the pretransport coop swabs. The prescald water samples were collected in 100-ml sterile specimen cups by removing approximately 100 ml of water from the scald tanks before birds entered the tank. The postscald water samples were collected in the same manner as the prescald after the birds had been run through the scald tanks. The prechill water samples were collected in specimen cups from the chill tank prior to birds entering the tank. The postchill water samples were pulled in the same manner after the birds had been removed from the tank. The carcass rinse samples were collected by placing a carcass into a Cryovac bag (Cat. no. B340; Cryovac, Duncan, S.C.), adding approximately 100 ml of sterile distilled water *(11),* vigorously shaking for 60 s, and pouring the rinse water into a sterile specimen cup. All samples were stored and shipped as previously described.

Sample preparation. Five hundred milliliters of BPW was added to each paper pad sample. The paper pads were kneaded and macerated by hand for 1 min. Five milliliters of BPW was added to all samples collected with cotton-tipped applicators. The samples were blended by stomaching for 30 s, and 1 ml of the sample was then placed into 9 ml of BPW. Ten milliliters of BPW was added to all samples collected with gauze pads or commercially prepared drag swabs. The samples were blended for 30 s, and 1 ml of the sample was placed into 9 ml of BPW. Ten grams each of all bulk samples was weighed out and placed into sterile specimen cups; 90 ml of BPW was added to the cup, and the sample was vortexed for approximately 15 s. Fecal droppings were weighed, then transferred to sterile specimen cups preenriched in 9 ml BPW/g feces. The water samples and carcass rinse samples were measured, and an appropriate amount of 10× BPW was added to bring the dilution to a 1× strength. All samples were then incubated overnight at 37°C.

Sample enrichment. After overnight preenrichment, 0.1 ml of the samples was transferred into 9 ml of tetrathionate broth, Hajna (Difco, Becton Dickinson) prepared according to the instructions on the packaging. The tetrathionate broth was incubated for 24 h at 42°C, and 0.1 ml was transferred into 9 ml of Rappaport-Vassiliadis broth (Difco, Becton Dickinson) prepared according to the instructions on the packaging. The Rappaport-Vassiliadis tubes were incubated overnight at 35°C.

Isolation and detection. The Rappaport-Vassiliadis enrichment tube samples were streaked on brilliant green (BG) sulfa (Difco, Becton Dickinson) with 15 ppm Novobiocin (Sigma Chemical Co., St. Louis, Mo.) added, xylose lactose tergitol 4 (XLT4; Difco Laboratories, Detroit, Mich.), and modified lysine iron agar (MLIA; Oxoid Inc., Ogdensburg, N.Y.) with 15 ppm Novobiocin agar plates. The plates were incubated overnight at 35°C. Two typical colonies were picked to triple sugar iron (Difco, Becton Dickinson) and lysine iron agar (Oxoid) slants, which were

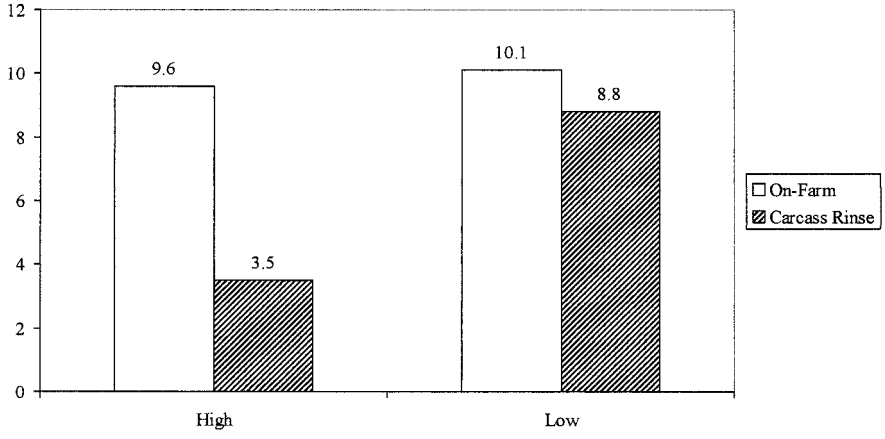

FIGURE 2. *Prevalence of* Salmonella *for all sample types from *high- and **low-production locations. * Top 25% of producers as measured by feed conversion and weight gain. ** Bottom 25% of producers as measured by feed conversion and weight gain.*

TABLE 4. *Serotypes of* Salmonella *from all sample types during growout, transport, and processing*

4,5, 12:I-Monophasic (27)[a]	4, 12:I-Monophasic (1)	4, 12:I-Nonmotile (1)
42:Z36 (1)	Agona (7)	Bareilly (1)
Braenderup (1)	Brandenberg (53)	Bredeney (1)
Derby (3)	Enteritidis (24)	Give (1)
Hadar (8)	Havana (1)	Heidelberg (50)
Infantis (40)	Inverness (2)	Johannesburg (1)
Kentucky (32)	Lille (2)	Livingstone (3)
Mbandaka (81)	Molade (6)	Montevideo (147)
Muenchen (1)	Newington (1)	Newport (4)
Ohio (8)	Ouakam (2)	Schwarzengrund (40)
Senftenberg (257)	Tennessee (5)	Thompson (208)
Typhimurium (22)	Typhimurium, Copenhagen (5)	Untypable (8)

[a] Frequency of isolation.

incubated at 35°C overnight. Samples giving typical reactions were serogrouped with *Salmonella* O antisera (Difco). The grouped cultures were transferred to duplicate Trypticase soy agar slants (Difco). The cultures were incubated overnight at 35°C. The tubes were sealed with Parafilm M (American National Can, Neenah, Wis.); one was stored in the laboratory, and the other was sent to the U.S. Department of Agriculture, Animal Plant Health Inspection Service, National Veterinary Services Laboratory, Ames, Iowa, for serotyping.

RESULTS AND DISCUSSION

A total of 10,740 samples from 20 on-farm and 6 transport or processing plant locations were tested for presence and serotype of *Salmonella*. For the four integrated operations, the sample types with the greatest frequency of *Salmonella* (Tables 1 and 2) were paper pads (50.8%), fly strips (18.7%), drag swabs (14.2%), and boot swabs (12%). All 26 sample types had some *Salmonella*-positive samples. Overall, on-farm samples were 9.8% positive for *Salmonella,* with a range from 5.2% for integrator A to 13.4% for integrator D (Table 1). The frequency of *Salmonella* recovery by sample time (Table 2) showed that *Salmonella* was recovered from most sample types throughout the entire grow-out period. As would be expected, the highest recovery rates were found in feces, litter, and drag swabs. For insects, flies, mice, and many other sample types, it is often difficult to determine if the chicken got the *Salmonella* from the sample or if the sample got the *Salmonella* from the chicken. For transport and processing plant samples (Table 3), the range of *Salmonella* recovery was from 2.0% for integrator C to 9.6% for integrator B. No *Salmonella* was recovered from prechill water samples, so these data were not included in Table 3.

The high frequency of recovery of *Salmonella* from flies (18.7%) is very interesting and suggests that flies may be an inexpensive and easy sample to screen houses and flocks for the presence of *Salmonella*. The high recovery rates of *Salmonella* from boot swabs (20 of 167, 12%) and the outside dirt (8 of 131, 6.1%) near the entrance doors to the houses show how easily movement and cross-contamination can occur and point out the need for an effective foot-bath system to help reduce this cross-contamination. Henzler and Opitz *(14)* and others have shown that in layer flocks, mice can harbor significant populations of *Salmonella* Enteritidis and may be a significant factor in the spread of this serovar. In this study, *Salmonella* was recovered from only 2 of 24 (8.3%) mouse rinses and 1 of 25 (4.0%) mouse entrails. The low recovery rates for mice suggest that the companies' rodent control programs were fairly effective and that the mice likely did not serve as the primary source of the *Salmonella* found on the processed carcasses. The recovery of *Salmonella* (10 of 731, 1.4%) from water lines suggests that under some conditions, *Salmonella* can form or be trapped in a biofilm layer in the water pipes or hoses.

Integrators participating in this study were from a wide geographical range; thus, the seasons were defined by months rather than by temperature. The prevalence of *Salmonella* by season is shown in Figure 1. For all seasons, a higher frequency of *Salmonella* recovered was found from on-farm samples compared to carcass rinse samples. Highest recovery rates were observed in the fall, followed by the winter, spring, and summer.

When high-production integrators were compared to low-production integrators (Fig. 2), there was very little difference in the recovery of *Salmonella* from on-farm samples. However, when carcass rinse samples were compared, there was a greater than twofold difference (8.8% positive carcasses from low-production farms and 3.5% positive carcasses from high-production farms) that is difficult to interpret. One possibility may be that even though there was little difference in the presence of *Salmonella* on the farm, the general health and condition that led to the increased feed conversion and weight gain of the high-production birds meant that there were lower levels of *Salmonella* present on these birds compared to those from the low-production houses. Recent studies *(5, 20)* have shown that when levels of *Salmonella* are low, the use of 40 to 50 ppm chlorine in the chill tank will reduce the rates of *Salmonella* coming out of the chill tank and processing plant.

From all sample types, there were 36 serotypes identified (Table 4). Twelve different serotypes were found on processed carcasses (Table 5). Of these, *Salmonella* Thompson was the most frequently identified serotype (29 isolates) followed by *Salmonella* Molade (4 isolates). Hatchery transport paper pads were the most frequently observed *Salmonella*-positive sample, and 9 of 12 serotypes found on

TABLE 5. *Serotypes found on the carcass and other sources during the growout*

Sample	4,5,12:I-Monophasic	42:Z36	Brandenberg	Infantis	Kentucky	Mbandaka	Molade	Montevideo	Ouakam	Semftenberg	Thompson	Typhimurium
Carcass rinse	1	1	1	4	1	1	4	2	2	2	29	1
Postchill water	0	0	1	0	0	1	1	0	0	2	0	0
Prescald water	0	0	0	0	2	0	0	0	0	1	0	0
Postscald water	3	0	0	0	2	0	0	0	0	0	0	2
Pretransport coop	0	0	0	3	0	3	1	0	0	3	1	0
Posttransport coop	0	0	5	2	6	1	0	2	0	3	5	1
Paper pads	4	0	9	28	18	45	0	38	0	194	53	7
Feces	4	0	16	0	2	8	0	39	0	16	58	0
Water line	0	0	1	0	0	0	0	0	0	6	1	0
Water cup	0	0	2	0	0	0	0	3	0	6	1	0
Litter	2	0	6	1	0	4	0	25	0	7	30	0
Feed hopper	0	0	0	0	0	0	0	0	0	0	1	1
Feeder	0	0	0	0	0	1	0	0	0	2	2	0
Drag swab	0	0	7	0	0	10	0	16	0	3	13	1
Wall swab	0	0	0	0	0	0	0	4	0	1	2	3
Fan swab	1	0	0	0	0	1	0	5	0	0	1	0
Mice sampling	0	0	0	0	0	0	0	1	0	0	0	0
Wild-bird feces	2	0	1	0	0	1	0	1	0	2	2	1
Animal feces	1	0	0	0	0	0	0	0	0	0	0	2
Insects	2	0	1	0	0	2	0	1	0	0	1	0
Dirt, near entrance	1	0	1	0	0	0	0	0	0	1	1	0
Standing water	0	0	0	0	0	1	0	1	0	0	0	0
Boot swab	1	0	1	0	0	2	0	5	0	1	1	2
Fly strip	5	0	0	1	1	0	0	1	0	6	1	0
Cecal droppings	0	0	1	1	0	0	0	3	0	1	5	1
Totals	27	1	53	40	32	81	6	147	2	257	208	22

processed carcasses were also found on paper pads. However, the most frequent serotype found on paper pads, *Salmonella* Senftenberg, was found primarily in one hatchery and only infrequently from on-farm samples during growout. A subsequent in-depth study of this hatchery showed that the *Salmonella* Senftenberg got on the paper pads in the hatchery through the air and did not usually result in colonized chicks. *Salmonella* Thompson was the second most frequently observed serotype from the hatchery and was frequently observed in feces, litter, and drag swab samples, indicating that significant colonization of chicks with this serotype likely occurred in the hatchery. *Salmonella* Molade was not found in the hatchery or in any on-farm samples, but it was found in the pretransport coop swabs, suggesting that the chickens likely picked up this serotype during transport from the farm to the processing plant. It is probable that the *Salmonella* Molade got into the transport coop from a previous flock that was being transported to the processing plant. These data are similar to observations by Rigby and Pettit *(18),* who found that chickens placed in transport crates with a marker strain of *Salmonella* became both carriers and shedders of that organism. Ten different serotypes were identified from feed samples; however, on only one occasion was the same serotype found in the feed also found on the final processed carcass. This observation is similar to that by Goren et al. *(13),* who found that serotypes of *Salmonella* found in the hatchery were much more likely to be found on processed carcasses than were serotypes found in feed.

Salmonella colonization of broiler chickens and subsequent spread through the integrated operation is complex. This multistate study examined all known potential sources of *Salmonella* across a full year's growout. Clearly, these data support the critical need to control *Salmonella* in the hatchery and in young chicks. Perhaps the most significant observation from this extensive survey is that *Salmonella* serotypes were recovered from all 26 different sample types. It is likely that no single intervention or *Salmonella* control strategy will consistently eliminate or significantly reduce *Salmonella* on the farm; therefore, multiple interventions to address different sources of *Salmonella* will need to be implemented to help control the entry and spread of *Salmonella* into the broiler operation from the almost ubiquitous presence of this organism throughout the industry.

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